ATTACHMENT B - CLEAN COPY OF SPECIFICATION

Please amend the specification at page 19, line 21:

Flagella are a key virulence determinant of Campylobacter spp. since motility is essential for establishment of colonization in the mucus lining of the gastrointestinal tract (25,26,27)

N/E

Please amend the specification at page 8, lines 13-21 through page 9, line 4:

Region I of the flaA gene represents the highly conserved N terminal region, and regions II and III represent two regions which are more variable among different sequenced flagellin genes. Regions II and III are not, however, as variable as region IV. The construct was made by amplifying the regions I, II and III using the primer flaA-11 (5'ACCAATATTAACACAAATGTTGCAGCA3') (Seq. ID no. 3) and flaA-2 (5'TTATCTAGACTAATCTCTACCATCATTTTTAAC3') (Seq. ID no. 4). The PCR product is digested with the appropriate restriction enzymes in order to insert the product into an expression vector. Any plasmid expression vector, e.g. PET™(Novogen, Madison Wisconsin) or PMAL™ (New England Biolabs, Beverly, MA) and viral expression vectors (e.g. adenovirus, M13, herpesvirus, vaccinia, baculovirus, etc) expression systems can be used as long as the polypeptide is able to be expressed. The PET™ vector is used for the cloning and over-expression of recombinant proteins in E.coli. In the PET™ system, the cloned gene is expressed under the control of a phage T7

B4

promotor. In the PMAL™ protein fusion and purification system, the cloned gene is inserted into a PMAL™ vector downstream from the MALE™ gene, which encodes maltose-binding protein (MBP). This results in the expression of an MBP-fusion protein. The technique uses the strong P_{tac} promotor and the translation initiation signals of MBP to express large amounts of the fusion protein. The PMAL-C2™ series of vectors have an exact deletion of the MALE™ signal sequence, resulting in cytoplasmic expression of the fusion protein. The PMAL-P2™ series of vectors contain the normal MALETM signal sequence, which directs the fusion protein through the cytoplasmic membrane, resulting in periplasmic expression. The preferred expression system is the PMAL-C2™ vector (New England Biolabs, Beverly, MA). For insertion into this system the PCR product is digested with SspI and XbaI, purified by agarose gel electrophoresis, and cloned in a commercially available plasmid vector, PMAL-P2™ or PMAL-C2™ (New England Biolabs, Beverly, MA) which had been digested with XmnI and XbaI. This vector allows for fusion of the fifth codon of the flaA gene to an *Escherichia coli* gene encoding maltose binding protein (MBP). The MBP-FlaA fusion is transcriptionally regulated by a P_{tac} promotor and is induced by growth in isopropylthiogalactoside (IPTG). Several transformants of *E. coli* DH5-alpha, containing plasmids with the appropriate size insert, were sequenced with the MALE™ primer (New England Biolabs). The MALE™ primer is used for sequencing downstream from the malE gene across the polylinker. One plasmid with the expected fusion-protein in the correct reading

B4

frame to [MalE]MALE™, termed pEB11-2, was purified.

Please amend the specification at page 9, line 11:

B5

Expression of recombinant flaA gene in PMAL-P2/C2™ plasmid.